

Claims

What is claimed is:

1. A modified protein comprising an amino acid sequence having an amino acid  
5 analog substituted at a specific amino acid residue, wherein lysine and/or cysteine side chains are  
not modified.

2. The modified protein of claim 1, wherein the amino acid analog is a tyrosine  
analog.

3. The modified protein of claim 2, wherein the tyrosine analog is acetyl-tyrosine.

4. The modified protein of claim 1, wherein the amino acid analog does not affect a  
biological activity of the protein.

5. The modified protein of claim 1, further comprising a label bound to the amino  
acid analog.

6. A method for producing modified proteins comprising the steps of:  
15 (a) synthesizing an amino acid analog, wherein the amino acid analog has  
selective reactivity; and

(b) incorporating the amino acid analog into a protein at a desired site,  
wherein the amino acid analog of the modified protein is capable of further modification.

7. The method of claim 6, wherein the further modification comprises the step of  
20 labeling the amino acid analog of the modified protein.

8. The method of claim 6, wherein the amino acid analog does not affect a biological  
activity of the protein.

9. A method for modifying proteins comprising:

- (a) identifying a protein having lysine or cysteine residues;
- (b) replacing an amino acid residue, other than lysine or cysteine, at a specific site in the protein with an amino acid analog.

10. The method of claim 9, wherein the amino acid analog does not affect a biological activity of the protein.

11. The method of claim 9, further comprising labeling the amino acid analog in the protein; wherein the label does not affect the biological activity of the protein.

12. The method of claim 9, wherein the protein is a Tat peptide (amino acids 47-56) (SEQ ID NO: 2).

13. The method of claim 12, wherein the Tyr-47 of the Tat peptide is the amino acid that is replaced.

14. The method of claim 13, wherein the amino acid analog is 3-Acetyl-Tyrosine.

15. A method for determining protein-RNA interactions under physiological conditions comprising the steps of:

- (a) labeling a site specific modified protein with a donor dye molecule, wherein the site specific modified protein comprises a protein modified by replacement of an amino acid with an analog of the amino acid, wherein the amino acid analog does not modify lysine or cysteine residues, and wherein the amino acid analog does not affect a biological activity of the protein;

- (b) labeling an RNA molecule with an acceptor dye molecule;
- (c) measuring the emission of the mixtures in step (a) and (b), respectively;
- (d) adding the mixture of step (b) to the mixture of step (a);

- (e) measuring the emission of the mixture in step (d); and
- (f) determining the interaction between the protein and an RNA molecule.

16. The method of claim 15, wherein the donor-acceptor dye pair is fluorescein-rhodamine.

5 17. The method of claim 15, wherein the site-specific modified protein is Acetyl-Tyr-Tat peptide.

18. The method of claim 17, wherein the RNA is TAR RNA.

19. A method for labeling proteins, without modifying lysine and cysteine side chains, comprising the steps of:

10 (a) replacing an amino acid of the protein, other than lysine and cysteine, with an analog of the amino acid; wherein the analog of the amino acid does not affect a biological activity of the protein; and

(b) labeling the amino acid analog of the protein with a dye; wherein the incorporation of the dye does not affect the biological activity of the protein.

15 20. A labeled protein comprising an amino acid sequence containing a plurality of lysine and/or cysteine residues, an amino acid analog, and a label located at the amino acid analog, wherein the amino acid analog and the label do not affect a biological activity of the protein.

21. The labeled protein of claim 20, wherein the amino acid analog is Acetyl-Tyrosine.

22. A method for producing site-specific modified proteins comprising the steps of:

(a) synthesizing an Acetyl-Tyrosine;

(b) incorporating the Acetyl-Tyrosine into a protein at a desired site, wherein the Acetyl-Tyrosine does not alter a biological activity of the protein, and wherein the Acetyl-Tyrosine is capable of further modification.

23. The method of claim 22, wherein the further modification comprises the step of  
5 labeling the Acetyl-Tyrosine of the site-specific modified protein.

24. A Tat peptide comprising an Acetyl-Tyrosine substituted for Tyrosine-47 in the Tat peptide (SEQ ID NO: 3), wherein lysine residues are not modified.

25. A labeled Tat peptide comprising a fluorescein-Acetyl-Tyrosine substituted for Tyrosine-47 in a Tat peptide.

26. A method for making the peptide of claim 24 comprising the steps of:  
10 (a) synthesizing an acetyl-tyrosine; and  
(b) synthesizing a Tat peptide, wherein the acetyl-tyrosine of step (a) is substituted for the Tyr-47 in the Tat peptide.

27. A method for making the peptide of claim 25 comprising the steps of:  
15 (a) synthesizing an acetyl-tyrosine;  
(b) synthesizing a Tat peptide, wherein the acetyl-tyrosine of step (a) is substituted for the Tyr-47 in the Tat peptide; and  
(c) site specifically labeling the acetyl-tyr-tat peptide at the location of the acetyl-tyr.  
20